# Supporting Information for

## Digital SERS-LFT Dipstick: Ultra-sensitive, Rapid Virus Quantification in Environmental Dust

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The Supporting Information contains 6 pages, 10 figures and 1 table.

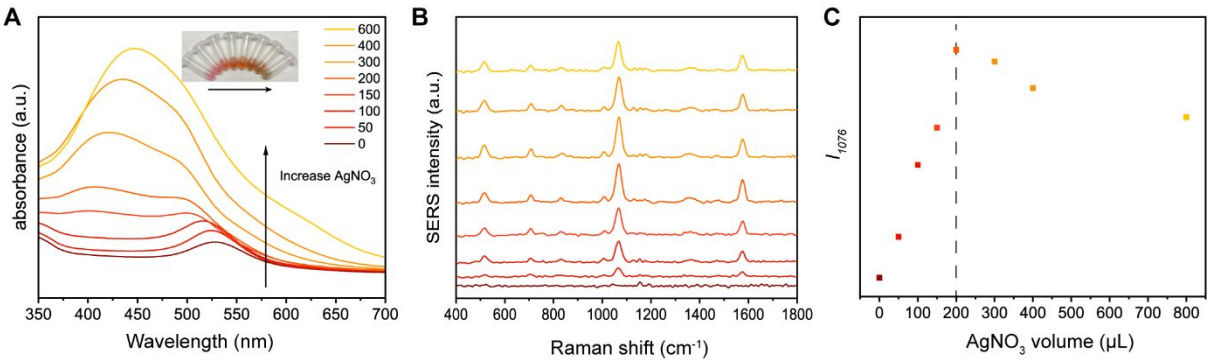


**Figure S1.** Optimization of shell thickness in core-shell $Au^{4\text{-}MBA}$@AgNPs by varying $AgNO_3$ dosage during synthesis. (A) UV-vis spectra; (B) Average SERS spectra obtained from different $Au^{4\text{-}MBA}$@AgNPs (C) The average peak intensity at 1076 $cm^{-1}$ obtained from different $Au^{4\text{-}MBA}$@AgNPs under different $AgNO_3$ dosage.

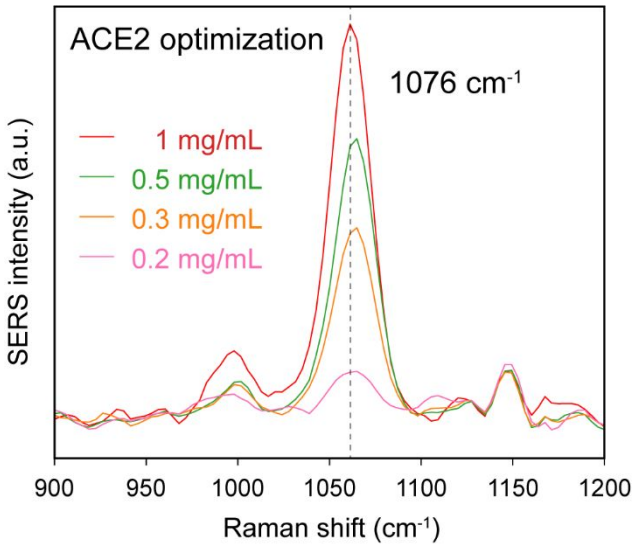


**Figure S2.** SERS intensity on the test dot during ACE2 concentration optimization using a dot-blot assay.

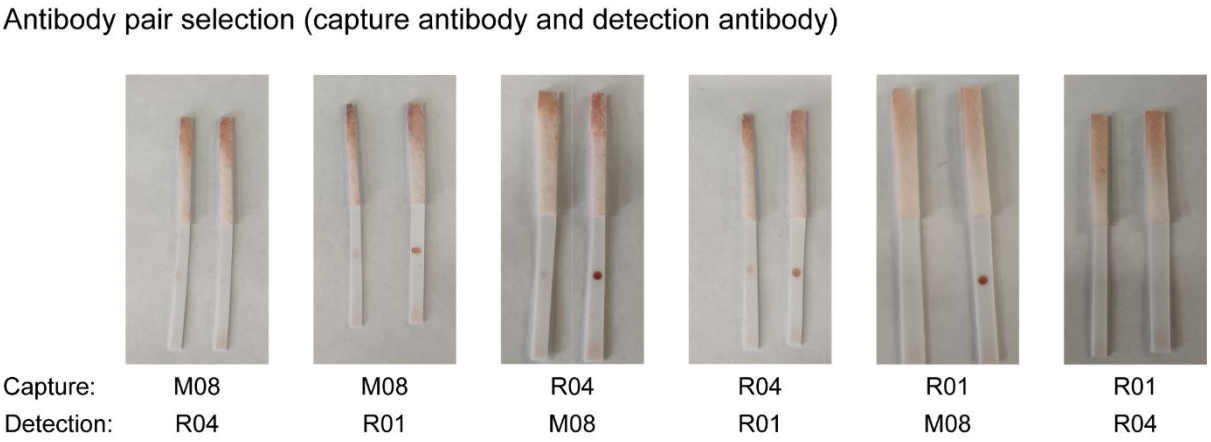


**Figure S3.** SERS-LFT strips using a dot-blot assay for nucleocapsid protein detection with different antibody combinations. The dots represent detection dots, and no control dots on the strips. The left strip in each photo refers to negative sample while the right strip represents positive sample.

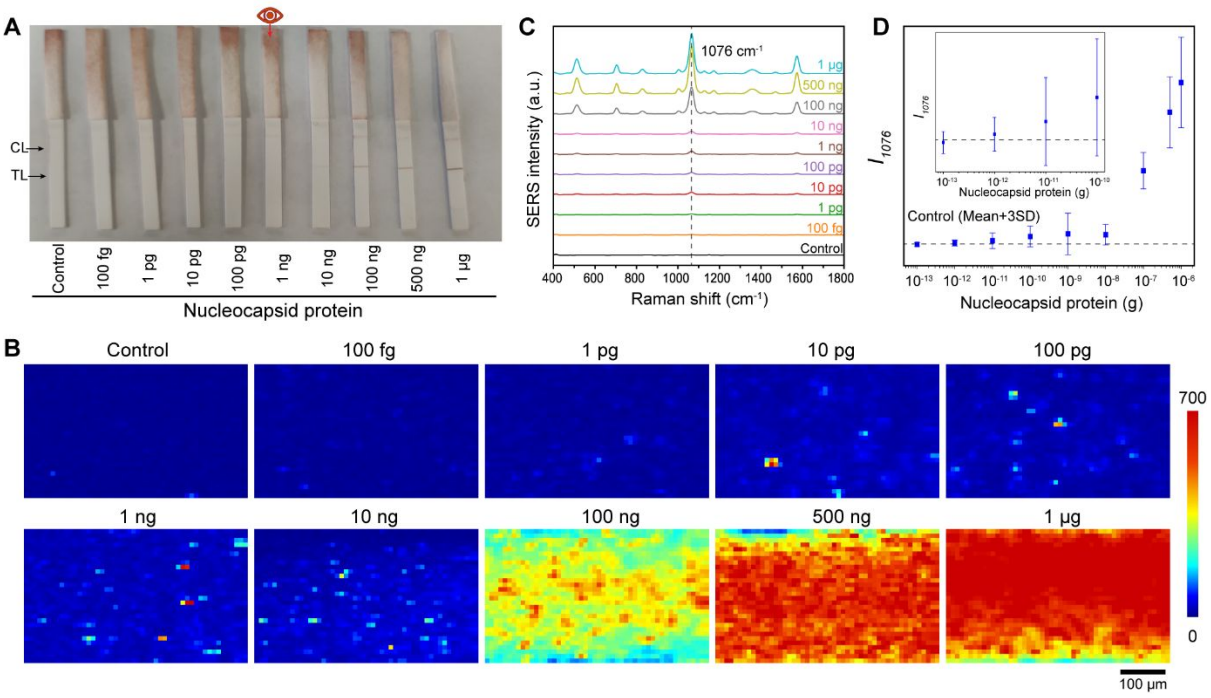


**Figure S4.** SERS- LFT for SARS-CoV-2 nucleocapsid protein detection. (A) Photograph of the SERS- LFT strips following the detection of nucleocapsid protein at different amounts; (B) SERS maps generated across the scanned area on the test line based on the peak intensity at 1076 $cm^{-1}$ ($I_{1076}$) for each sample; (C) Average SERS spectra obtained from the scanned area; (D) Comparison of $I_{1076}$ for each sample with control sample. The error bars represent the standard deviations obtained from area scans of each sample.

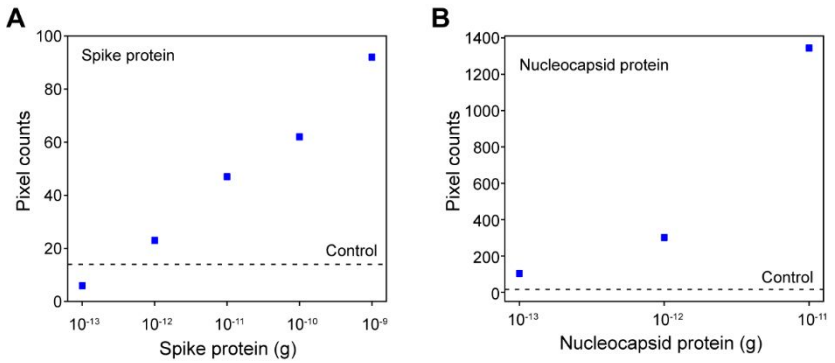


**Figure S5.** Pixel counts above the threshold for (A) spike protein and (B) nucleocapsid protein at different amounts. The dash lines indicate the pixel counts for control samples.

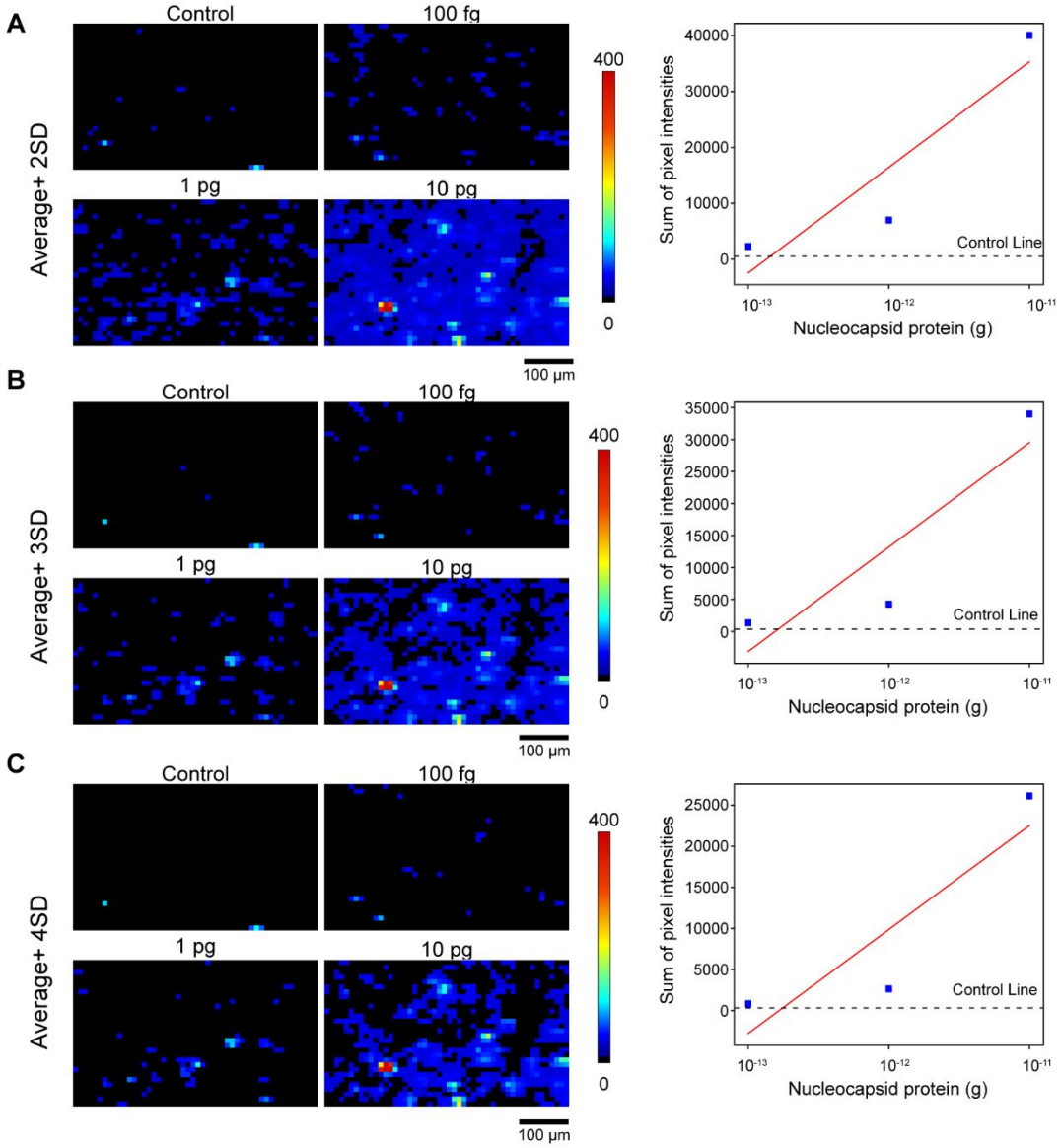


**Figure S6.** Digital SERS analysis for nucleocapsid protein quantification using different threshold values. (A) Average+2SD; (B) Average+3SD; (C)Average+4SD.

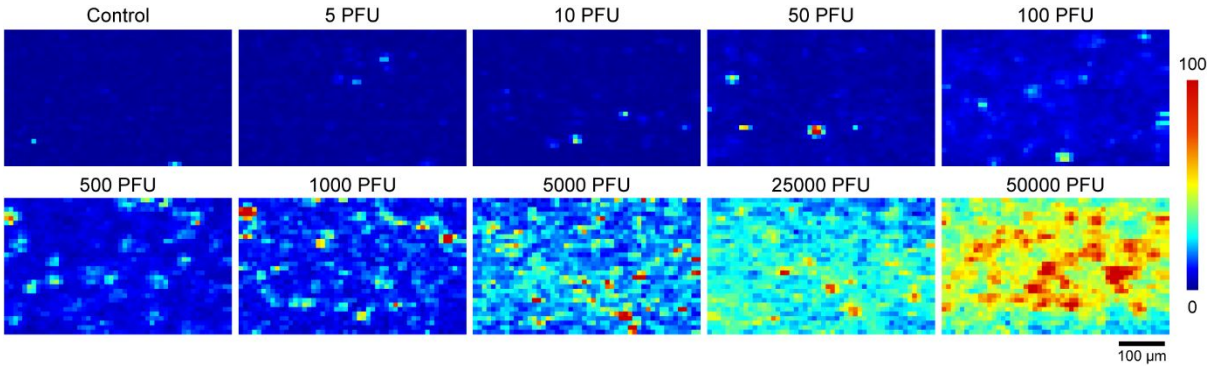


**Figure S7.** SERS maps generated across the scanned area on the test line for intact SARS-CoV-2 virus detection.

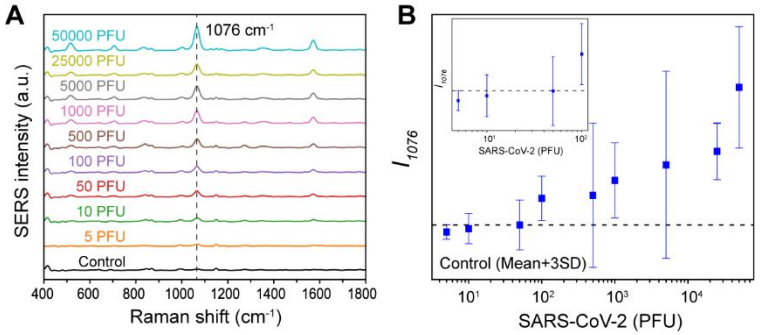


**Figure S8.** SERS- LFT for intact SARS-CoV-2 virus detection. (A) Average SERS spectra obtained from the scanned area; (B) Comparison of $I_{1076}$ for each sample with control sample. The error bars represent the standard deviations obtained from area scans of each sample.

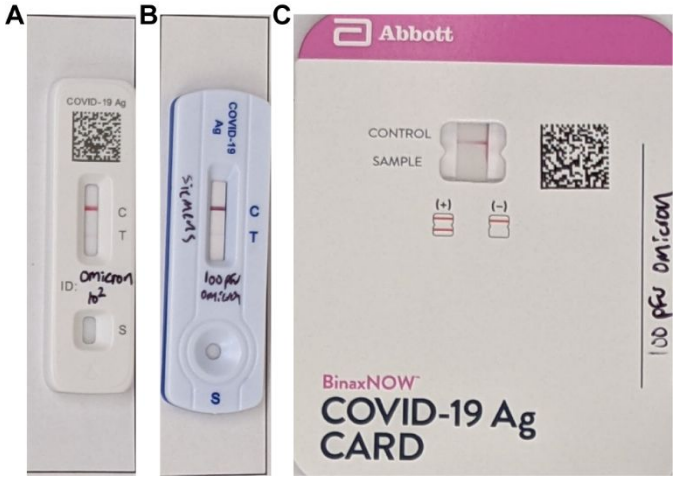


**Figure S9.** Intact SARS-CoV-2 virus detection using three commercially available antigen test kits at a concentration of 100 PFU. (A) FlowFlex; (B) Siemens; (C) Abbott.

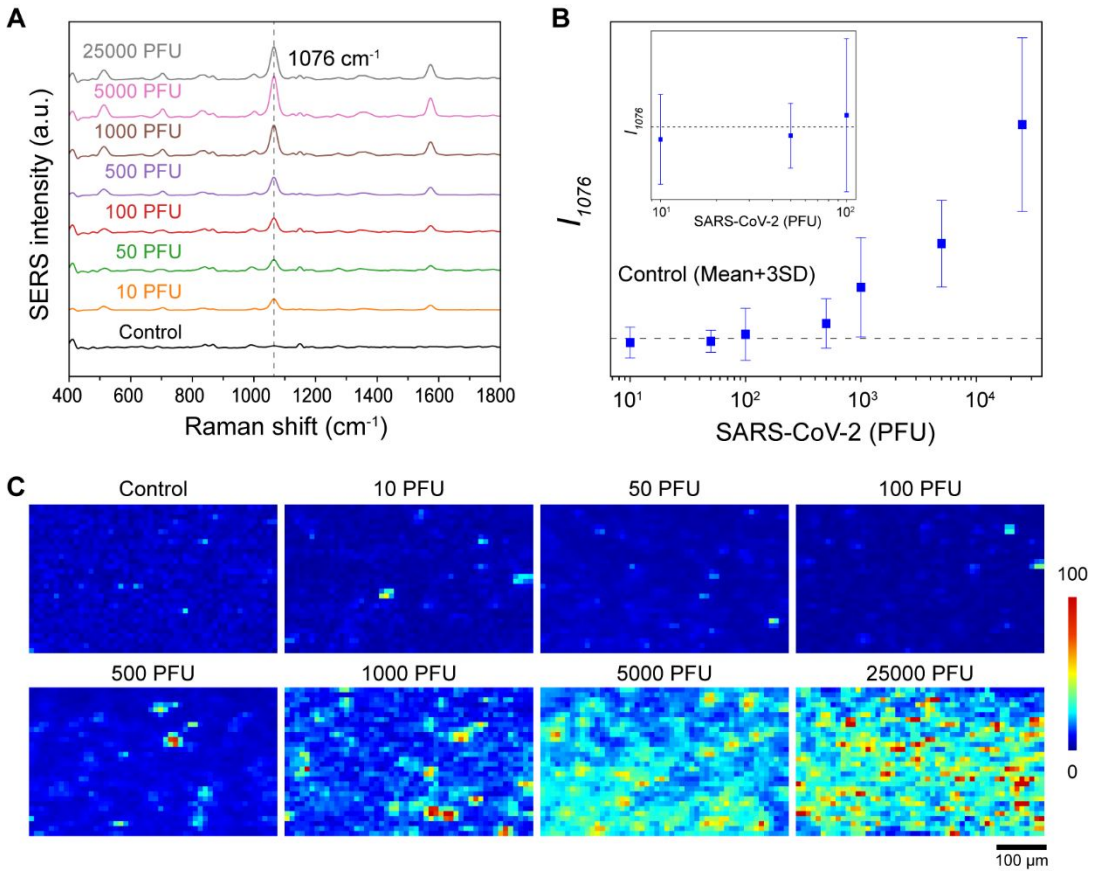


**Figure S10.** SERS- LFT for intact SARS-CoV-2 detection in indoor dust samples. (A) Average SERS spectra obtained from the scanned area; (B) Comparison of $I_{1076}$ for each sample with control sample. (C) SERS maps generated across the scanned area.

**Table S1.** Comparison of the detection performance of current digital SERS-based assays for SARS-CoV-2 proteins.

| Approach | Target | Matrix | LOD in concentration | Sample volume | LOD in mass | Assay time | Ref. |
|---|---|---|---|---|---|---|---|
| Digital SERS immunoassay | spike protein | PBS-10<br>saliva | 13.7 ng/mL<br>6.3 ng/mL | 30 μL<br>30 μL | 411 pg<br>189 pg | 5h | 1 |
| Digital SERS immunoassay | spike protein | / | 19 fg/mL | / | / | 5h | 2 |
| Digital SERS-LFT | spike protein<br>Nucleocapsid protein | PBS<br>PBS | 18 pg/mL<br>12 pg/mL | 10 μL<br>10 μL | 180 fg<br>120 fg | 30 min | This work |

**References:**

(1) Tuckmantel Bido, A.; Brolo, A. G., Digital SERS Protocol Using Au Nanoparticle-Based Extrinsic Raman Labels for the Determination of SARS-CoV-2 Spike Protein in Saliva Samples. *ACS Appl. Nano Mater.* **2023**, *6*, 15426-15436.

(2) Shim, J.-E.; Kim, Y. J.; Choe, J.-H.; Lee, T. G.; You, E.-A., Single-nanoparticle-based digital SERS sensing platform for the accurate quantitative detection of SARS-CoV-2. *ACS Appl. Mater. Interfaces* **2022**, *14*, 38459-38470.